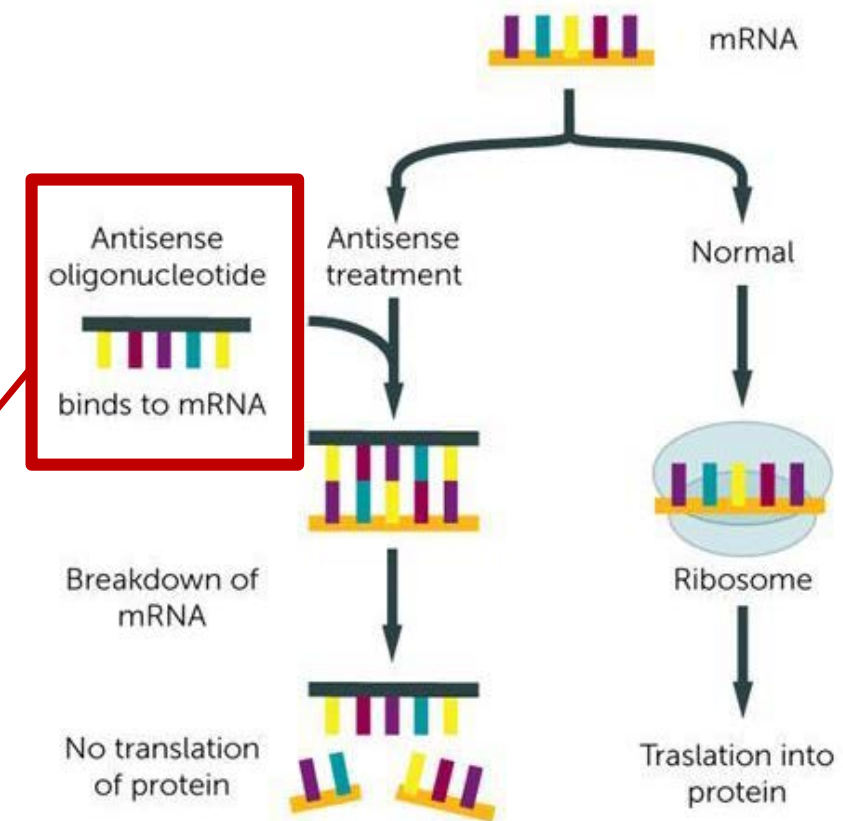

AUTOMATED DROPLET MANIPULATION THROUGH ELECTROWETTING ON DIELECTRIC FOR DNA SYNTHESIS

HEETAE AN ADVISOR: DR. MICHAEL SCHERTZER

WHAT IS AN OLIGONUCLEOTIDE?

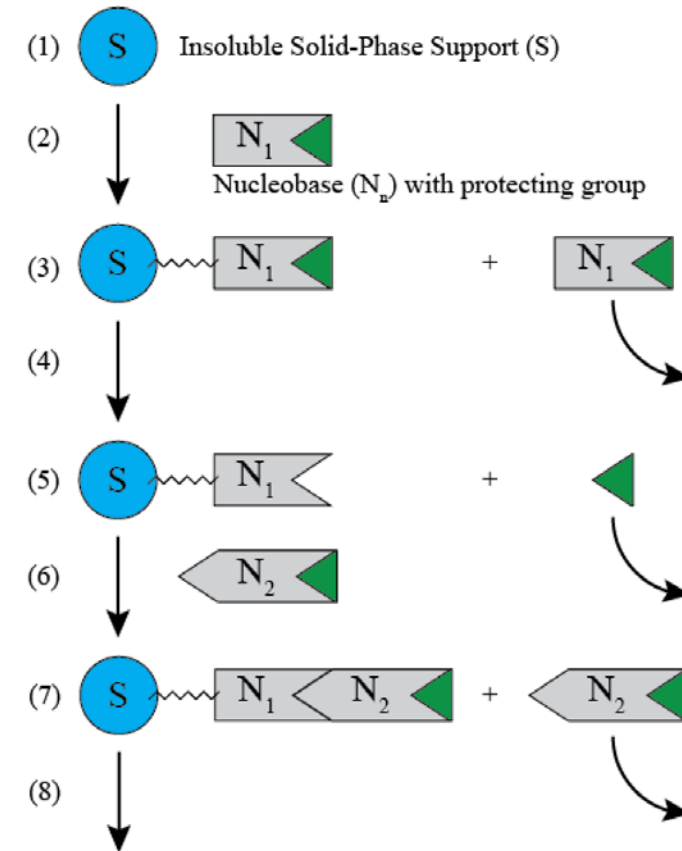
- Short DNA/RNA molecules
- Characterized by the sequence of nucleobases: adenine, guanine, cytosine, thymine, uracil
- Two use cases [1]:
 - Primers – Used to start chain reaction of target
 - Probes – Used to bind and hold target
- Common use is antisense therapy



Depiction of antisense oligonucleotide application [2].

SOLID-PHASE OLIGONUCLEOTIDE SYNTHESIS

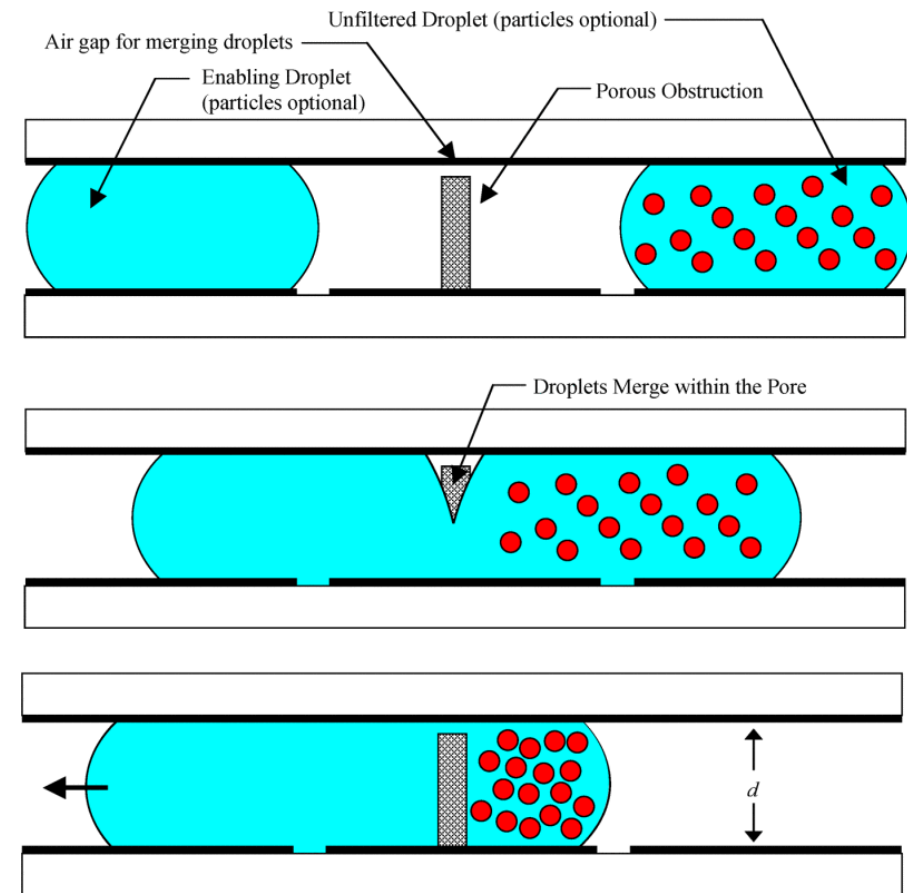
- 1) Addition of solid-phase support material
- 2) Addition of protected nucleobase
- 3) Attachment of protected nucleobase to solid support
- 4) Removal of unattached protected nucleobase molecules
- 5) Deprotection of attached nucleobase
- 6) Removal of detached protecting groups + addition of protected nucleobase
- 7) Attachment of protected nucleobase to deprotected nucleobase
- 8) Removal of unattached protected nucleobase molecules + reentry into cycle at step 4



Simplified depiction of solid-phase oligonucleotide synthesis [3].

WHY ELECTROWETTING ON DIELECTRIC (EWOD)?

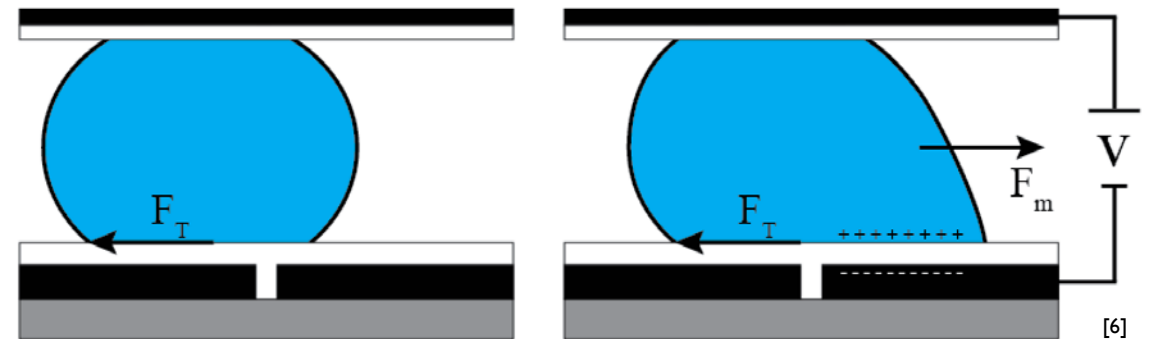
- EWOD allows for the manipulation of fluids
- Filtration techniques allows for selective removal of excess molecules
- Fluids can be split into droplets of very small volumes
- Devices can be designed to support many fluids simultaneously
- Manipulation can be easily automated



Depiction of particle filtration using SU-8 pillars [5]

EWOD ACTUATION THEORY

1. Voltage is applied to actuation electrode
2. Charge is induced in overlapping region of the droplet
3. Electrostatic force deforms the droplet, reducing the liquid solid surface tension at the leading edge
4. Surface tension gradient forces the droplet forward



$$F_m = \frac{\epsilon_0 \epsilon_R}{2d} V^2 - F_T$$

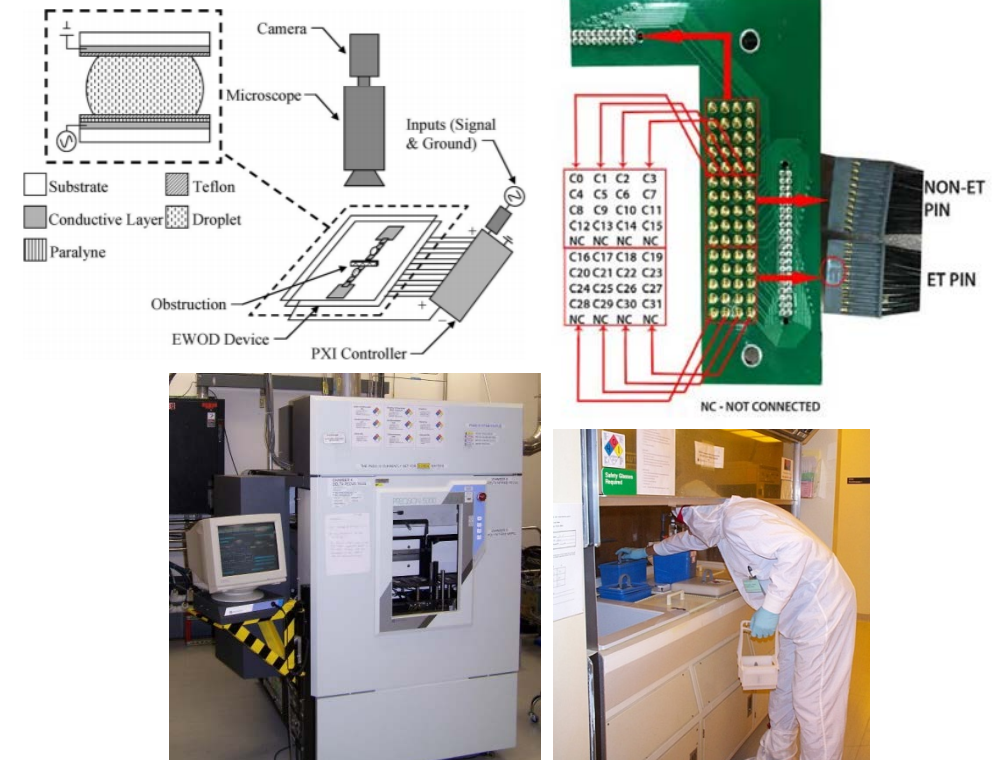
F_m : external force per unit length, ϵ_0 : permittivity of free space,
 ϵ_R : relative permittivity of the entire film stack,
 d : gap distance between the top and bottom electrodes,
 V : applied voltage, and F_T : threshold initiation force

PROJECT OBJECTIVES

- Design an electrowetting on dielectric (EWOD) device and process that manipulates droplets successfully, supports RIT SMFL fabrication processes, and also supports RIT DMFL testing facilities
- Perform and automate droplet manipulation with key functions such as droplet creation at reservoirs
- Perform and characterize particle filtration

DEVICE DESIGN CONSIDERATIONS

- Import from Schertzer et al. [5]
- RIT Semiconductor & Microsystems Manufacturing Laboratory (SMFL)
 - Wafer Standards
 - Available tools and chemistry
 - Supported chemistry
- RIT Discrete Microfluidics Laboratory (DMFL) testing facility
 - Pin placement and dimension
 - Mounting mechanism
 - Top plate ground



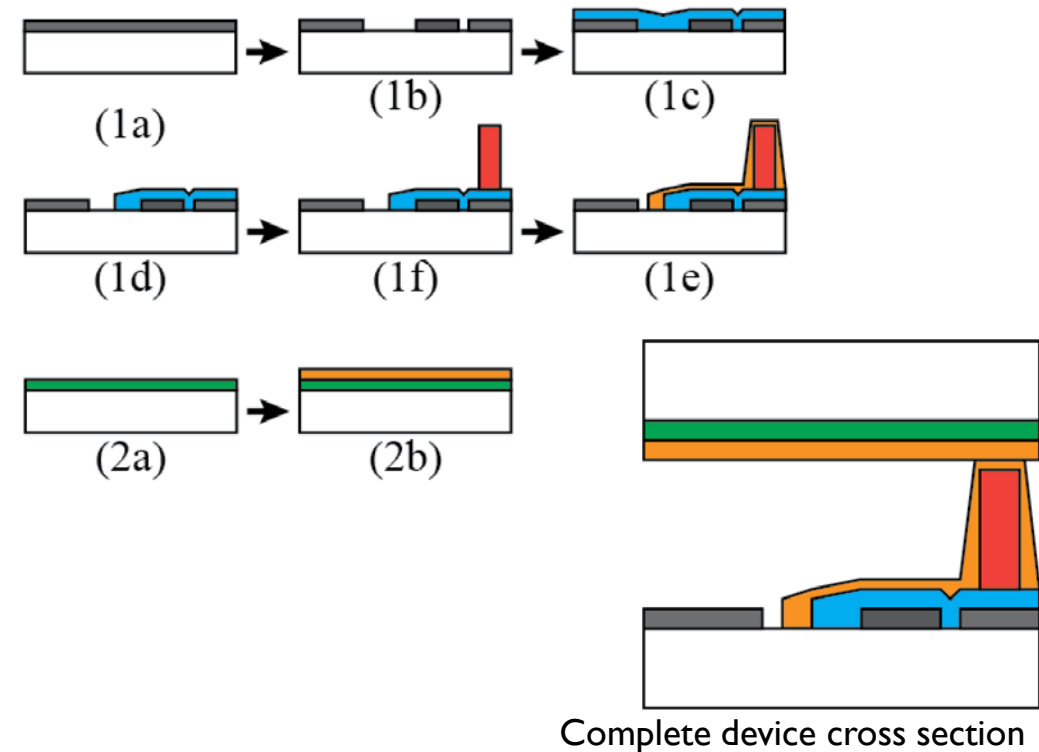
FABRICATION PROCESS FLOW

(1) Device Fabrication

- a. Al electrode PVD
- b. Electrode patterning
- c. SiO_2 insulator CVD
- d. Insulator patterning
- e. SU-8 filter lithography
- f. Teflon coating

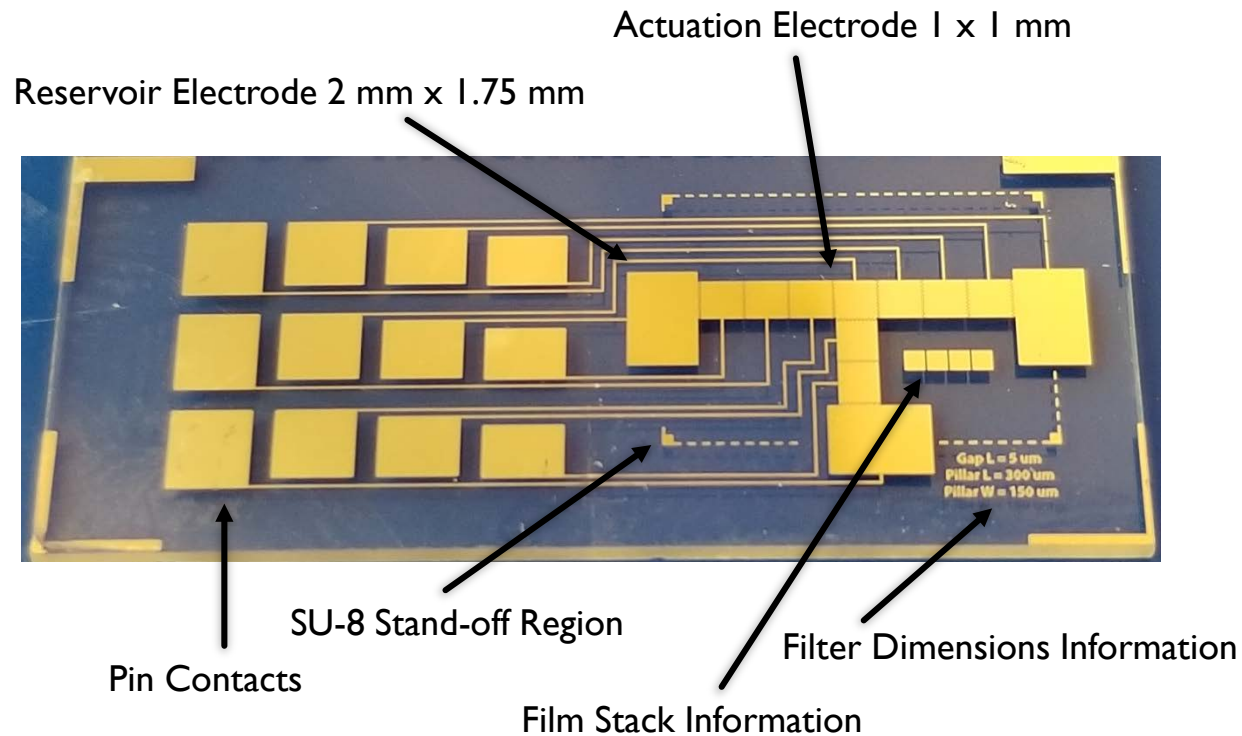
(2) Top Plate Fabrication

- a. ITO PVD
- b. Teflon coating

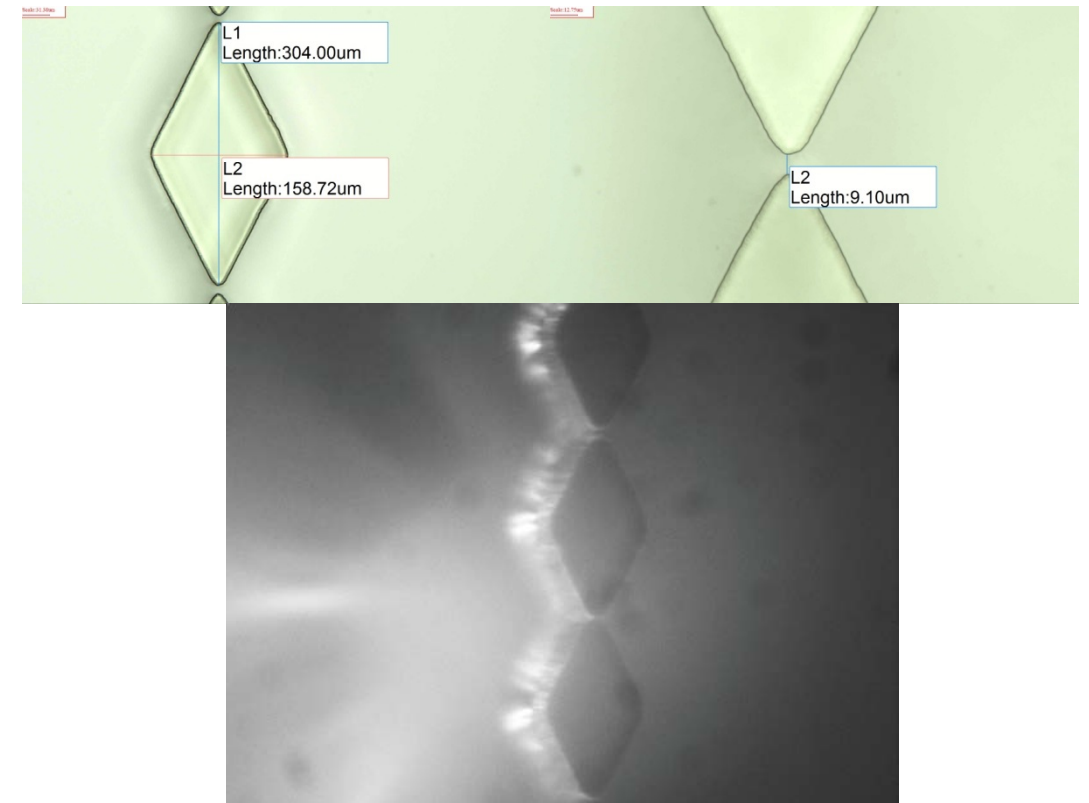


DEVICE FABRICATION RESULTS

SMFL Fabricated Device



SMFL Fabricated Filters on Device



AUTOMATED TESTING DESIGN

- a) Function generator signal control
- b) Digital multimeter
- c) Switch matrix initialization
- d) Program assignment and details
- e) Pin contact to pin assignment
- f) Split/merge and move timing with split/merge step number assignment
- g) Program status and control

The screenshot shows the Automated Testing Design software interface. Annotations (a) through (g) point to specific controls:

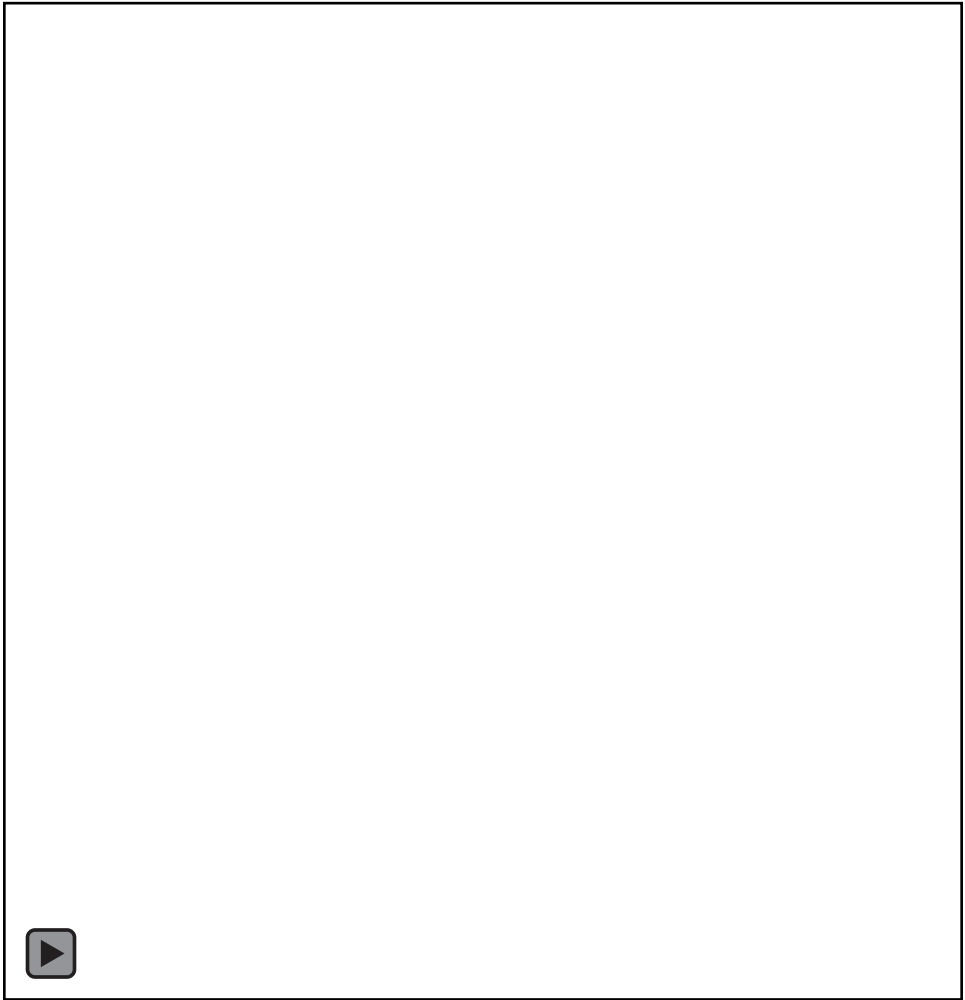
- (a) FGEN Device Name: PXI1 Slot 2
- (b) DMM Device Name: PXI1 Slot 4
- (c) SWITCH Device Name: PXI1 Slot 6
- (d) Program Name: R2R3.csv
- (e) Pin contact to pin assignment: R1 (c27), R2 (c20), R3 (c29), E1 (c26), E2 (c25), E3 (c28), E4 (c24), E5 (c21), E6 (c22), E7 (c23), E8 (c31), E9 (c30)
- (f) Split/Merge Time: 3
- (g) Program status and control: Status (Idle), OK, STOP/RESET

Current Program Details

	R1	R2	R3	E1	E2	E3	E4	E5	E6	E7	E8	E9
1	0	1	0	0	0	0	0	0	0	0	0	0
2	0	1	0	0	0	0	0	0	0	1	0	0
3	0	0	0	0	0	0	0	0	0	1	0	0
4	0	0	0	0	0	0	0	0	1	1	0	0
5	0	0	0	0	0	0	0	0	1	0	0	0
6	0	0	0	0	0	0	0	0	0	0	0	0
7	0	1	0	0	0	0	0	0	1	0	0	0
8	0	1	0	0	0	0	0	1	1	0	0	0
9	0	1	0	0	0	0	0	1	0	0	0	0
10	0	1	0	0	0	0	1	1	0	0	0	0
11	0	1	0	0	0	0	1	0	0	0	0	0
12	0	1	0	0	0	0	1	0	0	0	1	0
13	0	1	0	0	0	0	0	0	0	0	1	0
14	0	1	0	0	0	0	0	0	0	0	1	1

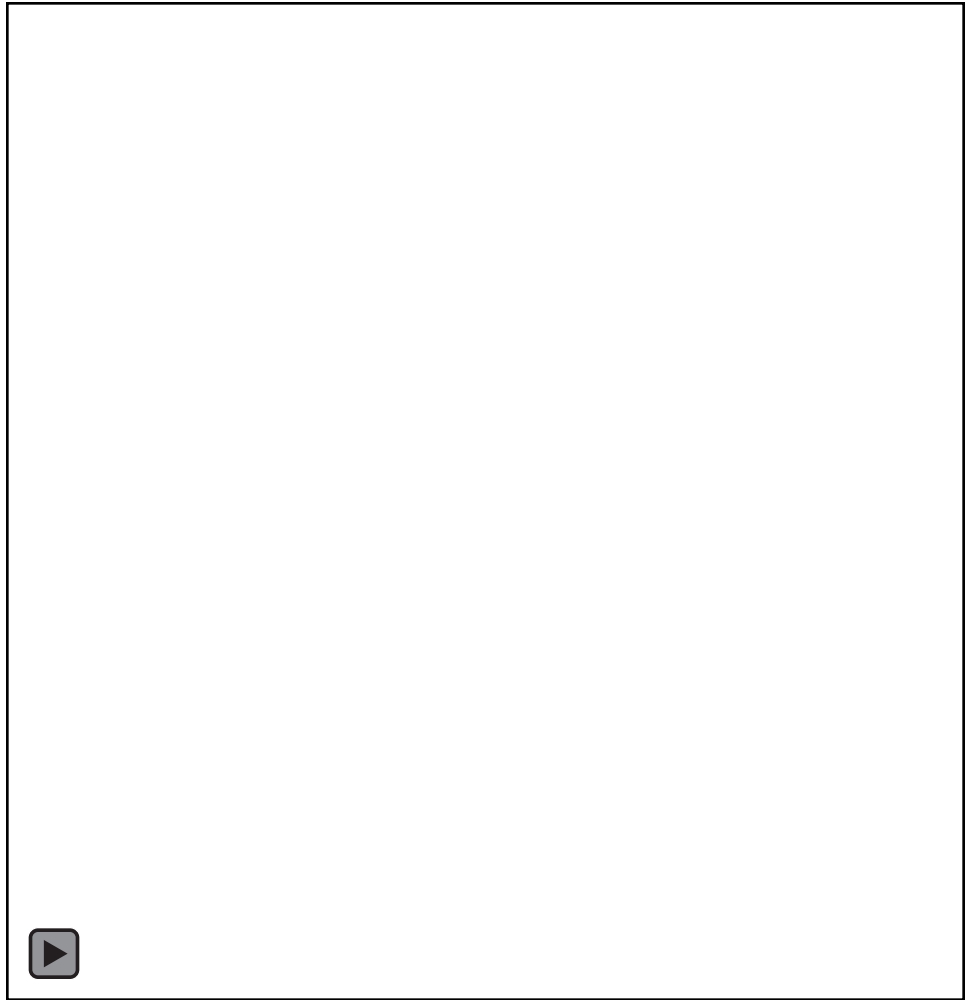
Current Step

0	0	0	0	0	0	0	0	0	1	0	0	0
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DROPLET MANIPULATION

35.5 Vrms, 1 kHz, 100-150 um gap



DROPLET MERGE/SPLIT

35.5 Vrms, 1 kHz, 100-150 um gap

CONCLUSIONS

- All the necessary device components for a complete DNA synthesis EWOD platform was designed, fabricated, and tested for functionality
- While the movement of droplets was easily performed, separation of droplets from reservoir seemed to be much more sensitive to the top plate distance
- Integration of the various device components will be necessary to perform reliable separation and allow solid-phase oligonucleotide synthesis
- Testing mounting changes will improve manipulation



QUESTIONS?





THANK YOU!

REFERENCES

- [1] “What is an oligo- or oligonucleotide?” [Online]. Available: <http://www.biosyn.com/faq/what-is-an-oligo-or-oligonucleotide.aspx>. [Accessed: 15-Apr-2018].
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- [5] Schertzer, M. J., R. Ben-Mrad, and Pierre E. Sullivan. "Mechanical filtration of particles in electrowetting on dielectric devices." *Journal of Microelectromechanical Systems* 20, no. 4 (2011): 1010-1015.
- [6] H. Ren, R. B. Fair, M. G. Pollack, and E. J. Shaughnessy, “Dynamics of electro-wetting droplet transport,” *Sensors and Actuators B: Chemical*, vol. 87, no. 1, pp. 201–206, Nov. 2002.